Sidransky and Baylin

Application No.: 10/659,519 Filing Date: September 9, 2003

Page 2

AMENDMENTS TO THE CLAIMS

Please amend claims 12, 13, 16, and 18 as set forth below. Claims 1-11 were previously canceled. Withdraw claims 20-24, without prejudice or disclaimer.

The current listing of claims replaces all prior listings.

1-11. (Canceled)

- 12. (Currently Amended) A method of detecting methylation of a p16 gene, comprising:
 - a) contacting a sample comprising nucleic acid molecules, with oligonucleotide primers that permit extension of a sequence complementary to amplification of a polynucleotide sequence encoding comprising exon 1 of the p16 gene and of a sequence complementary to a polynucleotide sequence encoding comprising exon 2 of the p16 gene, under conditions suitable for primer extension of the complementary sequences a nucleic acid amplification reaction; and
 - b) amplifying the resulting extension products of step (a) comprising contacting the extension products with a sense oligonucleotide which binds within and extends sequences from a 5' ALT promoter region; and
 - c) determining the presence of an amplification product that encodes a truncated p16 gene product, comprising detecting analyzing the amplification reaction for amplification products, wherein the presence of a[[n]] first amplification product comprising exon 2 of the p16 gene [[and]] in the absence of identifying a[[n]] second amplification product comprising exon 1 of the p16 gene, wherein hypermethylation of the 5' ALT promoter of the p16 gene is associated with the presence of the truncated product indicates the p16 gene is methylated, thereby detecting methylation of the p16 gene.
- 13. (Currently Amended) The method of claim 12, wherein step (a) further comprises contacting the sample with a demethylating agent, and wherein in the presence of the demethylating agent, the second product is detectable when methylation of the promoter results

Sidransky and Baylin

Application No.: 10/659,519

Filing Date: September 9, 2003

Page 3

in trucation of the p16 gene product the oligonucleotide primers that permit amplification of a polynucleotide comprising exon 2 of the p16 gene further permit amplification of polynucleotide comprising 5'ALT.

- 14. (Previously Presented) The method of claim 12, wherein the sample comprises a sample of a human.
- 15. (Previously Presented) The method of claim 12, wherein the sample comprises a biological fluid, cells, or a tissue.
- 16. (Currently Amended) The method of claim 13[[2]], wherein when the sample is contacted with the agent and the second amplification product is detected, methylation of the p16 gene is indicative of a neoplasm.
- 17. (Previously Presented) The method of claim 16, wherein the neoplasm is head and neck cancer, breast cancer, renal cancer, colon cancer, or prostate cancer.
- 18. (Curretnly Amended) The method of claim 12, wherein the <u>sample</u> nucleic acid molecules comprise RNA.
- 19. (Previously Presented) The method of claim 18, wherein the amplification reaction comprises reverse transcription and polymerase chain reaction.
- 20. (Withdrawn) A kit, comprising oligonucleotide primers that permit amplification of a polynucleotide comprising exon 1 of a p16 gene and exon 2 of the p16 gene.
- 21. (Withdrawn) The kit of claim 20, comprising a first forward primer that permits amplification of exon 1 of the p16 gene, a second forward primer that permits amplification of exon 2 of the p16 gene, and at least one reverse primer.

In re Application of:

Sidransky and Baylin

Application No.: 10/659,519

Filing Date: September 9, 2003

Page 4

22. (Withdrawn) The kit of claim 21, comprising one reverse primer, which permits amplification of exon 1 of the 16 gene and exon 2 of the p16 gene.

PATENT

Attorney Docket No. JHU1300-6

- 23. (Withdrawn) The kit of claim 21, comprising a first reverse primer that permits amplification of exon 1 of the p16 gene and a second reverse primer that permits amplification of exon 2 of the p16 gene.
- 24. (Withdrawn) The kit of claim 20, wherein the oligonucleotide primers that permit amplification of a polynucleotide comprising exon 2 of the p16 gene further permit amplification of a polynucleotide comprising 5'ALT.